

from the vegetation and from the livestock and reptile fauna. Among the reptiles (Tulcea county), *Testudo graeca iberica* (TGI) is a well represented species. Samples of ectoparasites obtained from TGI and livestock collected during the years: 2006–2007 and 2014–2015 (April–June) showed that the majority of ticks are represented by *Hyalomma aegyptium* and *H. marginatum*.

Over 400 *I. ricinus* and *Hyalomma sp.* ticks were collected and analyzed by Real-Time PCR methods (including the new TickItqPCR detection concept; project funded by the MEN-UEFISCDI PN II “Partnerships in priority areas” program, National Research Grant No. 295/2014”) that give us results on *Borrelia burgdorferi*, *Francisella tularensis*, TBEv and CCHFv (BFTC) presence in the vectors.

**Methods & Materials:** Total RNA and DNA were extracted and analyzed by in house real-time PCR reagents (included in the new TickItqPCR detection concept) and 2 commercial kits for the BFTC detection in the *I. ricinus* and *Hyalomma sp.* pools.

**Results:** Specific DNAs from *B. burgdorferi* *sl.* were detected (Fla B gene target) in 20% of *I. ricinus* ticks and specific DNAs for *F. tularensis* were detected (IS Ft2 genomic insertion-like element) in 2% of the same vector species.

Specific RNAs from TBEv were detected (3' UTR-genomic region) in < 1% of *I. ricinus* pools.

No specific CCHFv RNAs were detected (S –genomic region) among the *Hyalomma sp.* pools.

**Conclusion:** The results strengthen the concern that already exists in Romania, for the enhancement the surveillance and the control measures for the tick populations but also for the means of active information of the human population about the danger of the diseases transmitted by ticks in some risk areas.

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Date: Thursday, March 3, 2016

Time: 12:45–14:15

Room: Hall 3 (Posters & Exhibition)

#### SPIDR-WEB: an NGS biotechnology platform for diagnostic and transcriptomic applications



A. Hatch, P. Chain, J. Gans, M. Vuyisich\*

Los Alamos National Lab, Los Alamos, USA

**Background:** We are transforming the field of infectious disease diagnostics with the development of the Sample Prep for Infectious Disease Recognition With EDGE Bioinformatics (SPIDR-WEB). SPIDR-WEB is a sample-to-result biotechnology platform that enables efficient use of next generation sequencing (NGS) for pathogen detection in clinical samples. NGS has become a powerful tool for detection and characterization of both known and emerging pathogens. The main advantage of NGS is its non-biased approach that identifies all organisms in a sample. This is in contrast to traditional molecular assays that force us to look for a set of specific pathogens. In most clinical samples, the relative abundance of pathogen nucleic acids (DNA or RNA) is vanishingly small. Therefore, vast amounts of sequence data must be generated and analyzed to identify rare pathogen sequences. SPIDR-WEB is a sample-to-result process that relies on efficient laboratory and *in silico* steps.

**Methods & Materials:** Clinical samples mostly comprise non-informative host RNAs or abundant housekeeping gene transcripts. SPIDR-WEB incorporates removal of non-informative RNAs (RNR),

thereby enriching all other RNAs, including those from pathogens. This step enables either higher sensitivity and specificity, or less expensive and faster sequencing. Our custom EDGE bioinformatics data analysis platform provides rapid read classification at all taxonomic levels, and reliably detects all organisms present in a sample. EDGE is an efficient process, as it uses databases with pre-computed signatures, instead of aligning sequencing reads to the entire Genbank. In addition to RNR and EDGE, SPIDR-WEB includes robust, inexpensive and rapid sample lysis, RNA extraction, and library preparation steps.

**Results:** We will describe SPIDR-WEB technology and show clinically-relevant results obtained from human blood, stool, respiratory, and other sample types.

**Conclusion:** We are implementing SPIDR-WEB in both research and clinical settings to support a multitude of applications, such as discovery of novel mechanisms and biomarkers, study host-pathogen interactions, improve vaccines and therapeutics, and complement current diagnostic tools and help improve their utility.

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#### Yellow fever threat to Asia: A model national contingency plan



J. Woodall

Federal University of Rio de Janeiro, Rio de Janeiro, Brazil

**Background:** In December 2015, the Hong Kong Standard reported that its hospitals were ready to handle dengue and yellow fever outbreaks. There has never been a case of yellow fever in HK, or anywhere else in Asia. Nobody knows why not. But the risk is there. HK could lead Asia by preparing a contingency plan. Why? Because when YF arrives in HK everyone will think it is only jaundice or dengue.

**Methods & Materials:** Do the medical labs have the reagents to distinguish yellow fever from closely related dengue? Is there a stockpile of the vaccine? There is no treatment. Yellow fever kills 20% of unvaccinated people, the vaccine is only fully effective 10 days after the shot, and infants under 9 months and seniors over 65 should not have it because of possible side effects. Is there a stockpile of mosquito control chemicals, ground & air fogging equipment and technicians who know how to calibrate the spray and get themselves tested for the side effects of their exposure to the spray?

**Results:** A PowerPoint slide show of how to prepare a contingency plan is available from WHO/SEARO New Delhi.

**Conclusion:** We urge all Asian countries at risk – particularly India & southern China – to draft a contingency plan as soon as possible. This also applies to every country in Asia that has dengue, because the same mosquitoes that transmit dengue can transmit YF from an imported case. This is to avoid falling into the same unprepared situation that West Africa experienced with Ebola.

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John “Jack” Woodall PhD was a consultant for the WHO/SEARO 2011 Goa workshop on yellow fever.  
Rio de Janeiro, Brazil

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#### Eating raw liver, a potential risk factor of Crimean-Congo hemorrhagic fever (CCHF) occurrence in high-risk occupations in Nur County, Northern Iran

S.P. Ziapour<sup>1,\*</sup>, A. Enayati<sup>1</sup>, S.H. Nikookar<sup>1</sup>, F. Sahraei-Rostami<sup>1</sup>, S. Kheiri<sup>2</sup>, A. Charkameh<sup>1</sup>, J. Shojaei<sup>1</sup>

<sup>1</sup> Mazandaran University of Medical Sciences, Sari, Iran, Islamic Republic of

<sup>2</sup> Pasteur Institute of Iran, Sari, Iran, Islamic Republic of



**Background:** Eating raw lamb or beef liver is of interest to some people such as livestock farmers, shepherds, butchers and chefs. This high risk practice can cause gastrointestinal problems such as parasitic diseases and microbial infection of the liver tissue and also causes transmission of Crimean-Congo Hemorrhagic Fever (CCHF) to humans. CCHF cases occurrence in a butcher and a farmer in Nur County, raised the necessity to check the background of high risk behavior related to the consumption of raw liver in individuals who work in livestock and meat industry. This can help prevention of the disease in high-risk occupation groups.

**Methods & Materials:** In 2012, a cross-sectional study on 314 people, including livestock farmers, butchers, abattoir workers, chefs and veterinary staff was undertaken in three district of the Nur County. The practice of eating raw liver in different high risk occupation groups was recorded in a standard questionnaire through interview. The relevance of this high risk behavior with qualitative variables by Chi-square test and binary logistic regression were analyzed at the significant level of 0.05.

**Results:** The odds ratio (OR) of raw liver eating was significantly higher in livestock farmers and animal keepers than other occupations (OR = 15.27, CI: 2.04–114.32), in the mountains than plains and woodland areas (OR = 3.47, CI: 1.66–7.29) and in Baladeh district than other districts including Central and Chamestan District of the County (OR = 2.49, CI: 1.14–5.42), respectively. Additionally, consumption of raw liver in 30–39 year old age group was higher than other age groups (OR = 2.06, CI: 0.81–5.22), it was higher in rural population than urban residents (OR = 1.79, CI: 0.72–4.46). The prevalence of this behavior in mountainous areas and Baladeh District of Nur County may be explained by the high frequency of traditional animal husbandry and unsafe slaughtering, low literacy levels, difficulty in implementing policies on health education in rural areas away from the County.

**Conclusion:** People attempt to eat raw liver in high risk occupational groups can increase the risk of CCHF and even its epidemic in the region.

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#### Outbreak of *Prototheca wickerhamii* algaemia and sepsis in a tertiary care chemotherapy oncology unit



I.D. Khan

CH EC Kolkata, Kolkata, West Bengal, India

**Background:** *Prototheca* is an emerging, rare, opportunistic, pathogenic, achlorophyllous green alga known to cause Protothecosis which is a zoonotic disease. Earlier interpreted as contaminants in blood and faeces, *Prototheca* is expanding its pathogenicity and host range. An outbreak of Protothecosis by *Prototheca wickerhamii* in a tertiary care chemotherapy oncology unit is being discussed.

**Methods & Materials:** All patients detected to have algaemia were operationally included in the case definition. Clinicodemographic profile, diagnosis, duration of stay, treatment protocol and neutrophil count were correlated. After isolation on sheep blood and Sabouraud's agars, urease, Germ tube formation and automated identification through VITEK 2 (bioMérieux, France) were attempted. Colony characteristics, micromorphology, substrate utilization and antifungal susceptibility were interpreted. All patients were initiated on liposomal amphotericin B (5 mg/kg body weight/day). Fecal cultures of affected patients, environmental surveillance and healthcare staff were screened while continuing surveillance for one year post outbreak.

**Results:** The outbreak lasted approximately 50 days during which the average occupancy was 26 patients (86.67%) and mean hospital stay was 60 days. Mean age of affected patients was 37 ± 10.74 years with male: female:: 5: 1. Mean neutrophil count in affected patients was 150 per dl. The attack rate was 7.69. *Prototheca wickerhamii* was isolated on sheep blood and Sabouraud's agars as yeast-like colonies having Gram positive 3–11 µ non-capsulated spherical yeast-like cells without budding and pseudohyphae. All isolates were negative for urease and Germ tube formation. VITEK 2 compact provided 99% identification probability. MICs in µg/ml for Amphotericin B and Voriconazole were 0.5 and 2 respectively. All isolates were similar for biochemical reactions and susceptibility patterns. All patients responded to liposomal amphotericin B. One patient detected to have algaemia went into sepsis with serum procalcitonin levels between 2–4 ng/ml with subsequent fatal outcome under intensive care. Surveillance studies were not contributory.

**Conclusion:** Immunocompromised neutropenic patients having Protothecosis may not manifest clinical features leaving detection to intuitive clinical acumen. Outbreaks are difficult to detect and control as incubation period is variable. Such hospital outbreaks re-emphasize the need to strengthen hospital and laboratory based surveillance to ensure adequate preparedness, rapid detection and response to outbreaks.

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